

D0128 NP

Figure 1A

1 CCACGCGTCCGCTCAGGTCTATAGGATTAAGAAAGGCAAGCCCAGCAGCAGCTACTCACT 60
 61 GACCAGACCTGGCCCAACATGCTGCAGAAATAATTATCAATTAGTATACTTGAGAGACAG 120
 121 CAGCGTGAGGTGGAGAATGGGTTCTAACTGAATGACAGCTGTTAACAGTTTTGGCCCT 180
 181 GTTTTTCCTGCTCTGAATCTCAACTGAGATCCTAGGGATGAGAAACGGGGGAACAGCAC 240
 241 GCCCTACTTGAGAGAATTAGAATTTGAGGCGCTAGGAAGCAAAAGGATCCCAAAGATGGC 300
 301 GACCTGCCAGCCTGGACTGCCAGCGAAGGCCAGAATCGTGCTGTAGCTCTGAACCCACAG 360
 361 CTCCTCTGCCCTTGCCCATGAGAATTTCACTGGAGAGATAGCATGCCCTGGTAAGTGA 420
 421 AGTCTGCCACTTCGAGACATGGAATCATCTTTCTCATTGGAGTAGCTTGCTGTCTCT 480
 1 M E S S F S F G V I L A V L 14
 481 GGCCTCCCTCATCATTTGCTACTAACACACTAGTGGCTGTGGCTGTGCTGTGTTGATCCA 540
 15 A S L I I A T N T L V A V A V L L I H 34
 541 CAAGAATGATGGTGTGCTCTCTGCTTCACCTTGAATCTGGCTGTGGCTGACACCTTGAT 600
 35 K N D G V S L C F T L N L A V A D T L I 54
 601 TGGTGTGGCCATCTCTGGCCTACTCAGACAGCTCTCCAGCCCTTCTCGGCCACACA 660
 55 G V A I S G L L T D Q L S S P S R P T Q 74
 661 GAAGACCTGTGCGACCTGCGGATGGCATTGTCACTTCTCCGAGCTGCCTCTGTCTCT 720
 75 K T L G S L R M A F V T S S A A A S V L 94
 721 CACGGTCATGCTGATCACCTTTGACAGGTACCTTGCCATCAAGCAGCCCTTCCGCTACTT 780
 95 T V M L I T F D R Y L A I K Q P F R Y L 114
 781 GAAGATCATGAGTGGGTTTCGTGGCCGGGGCCTGCATTGCCGGGCTGTGGTTAGTGTCTTA 840
 115 K I M S G F V A G A C I A G L W L V S Y 134
 841 CCTCATGGCTTCTCTCCACTCGGAATCCCATGTTCCAGCAGACTGCCTACAAAGGGCA 900
 135 L I G F L P L G I P M F Q Q T A Y K G Q 154
 901 GTGCAGCTTCTTGCTGTATTTACCCCTCACTTCGTGCTGACCCCTCTCCTGCGTTGGCTT 960
 155 C S F F A V F H P H F V L T L S C V G F 174

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Figure 1B

961	CTTCCAGCCATGCTCCTCTTTGTCTTCTTCTACTGCGACATGCTCAAGATTGCCTCCAT	1020
175	<u>F P A M L L F V F F Y</u> C D M L K I A S M	194
1021	GCACAGCCAGCAGATTTCGAAAGATGGAACATGCAGGAGCCATGGCTGGAGGTATTCGATC	1080
195	H S Q Q I R K M E H A G A M A G G Y R S	214
1081	CCCACGGACTCCCAGCGACTTCAAAGCTCTCCGTACTGTGTCTGTCTCATTTGGGAGCTT	1140
215	P R T P S D F K A L R <u>T V S V L I G S F</u>	234
1141	TGCTCTATCCTGGACCCCTTCTTATCACTGGCATTGTGCAGGTGGCCTGCCAGGAGTG	1200
235	<u>A L S W T P F L I T G I V</u> Q V A C Q E C	254
1201	TCACCTCTACTAGTGTGGAACGGTACCTGTGGCTGCTCGGCGTGGCAACTCCCTGTCT	1260
255	H L Y L V L E R <u>Y L W L L G V G N S L L</u>	274
1261	CAACCCACTCATCTATGCTTATGGCAGAAGGAGGTGCGACTCGAGCTCTACCAATGGC	1320
275	<u>N P L I Y A Y W</u> Q K E V R L Q L Y H M A	294
1321	CCTAGGAGTGAAGAAGTGCTCACCTCATCTCTCTCTTTCTCTCGGCCAGGAATTGTGG	1380
295	L G V K K V L T S F L L F L S A R N C G	314
1381	CCCAGAGAGCCCGAGGAAAGTTCCTGTCACATCGTCACTATCTCCAGCTCAGAGTTTGA	1440
315	P E R P R E S S C H I V T I S S S E F D	334
1441	TGGCTAAGACGTGCTTTGCTTACAGTCTGGCCCCAGGAGAGAAACATGCTTGCTTTAC	1500
335	G	335
1501	CATAGCAAGCATCGTGTCTCTACAACCTGAAGAACTTTTGA	1560
1561	ACCCAATATGCCTTGAAATGCTGAGCATCCAGCAAGTACCAGGCCAGCAATAAGGCACAT	1620
1621	TCTTGGATTGTATTGTGAGTTACTGGGAAC	1680
1681	CAAAATGAAAGCAGGCTCAAACCAATGCCTCCTTACTGGTGGTTCAAAGCTTAAATCC	1740
1741	TGGCCTTGCTACAAAGGACTGATCTTGCCAAGATAATATAAGCAAAGTGAATGAAATT	1800
1801	AAAGTTAATTCTGAAGCCAAGGTCCTTTTAGAAAAAAAAAAGTAAATTAGCCTGATTTAA	1860
1861	TCATTCCACATTTTAAACATATATCAACATCATTGTACCCACGTAATATATACAATT	1920
1921	ATTTGTCAATTAAAAATAAAAAATATATATTTTAAAGTACAGAAAAGCAGGAGGGAGGA	1980

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Figure 1C

1981 GAGGCCAGGTGATCAGTGTGTTATTAGGTTCAATCTAAGAACTCTGCTACCCCTGGGAG 2040
2041 TCAGTATGTGCAATGAAAAATCACAGGTCTTGAAATCCAACAGACCAGTGTTCAAATGA 2100
2101 TAATTCAGTCATTTAAATTCTCAAAC TAGATCTCGCATT CATCAAACCGGGTAATAATG 2160
2161 CCTACTTTACATGGTTATATTGAAGATTAAC TCAGATAATGTATATGTAAATATCTAGTA 2220
2221 AACTACAGCACATTGTTAGTGCTCAAAAAAAAAAAAAAG 2260

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Figure 2A

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B2AR_CANFA -----MCQPANRSVFLAPNCSAPPDQDSQRSEAMV.VGAILVMSVLIVLAIVPNNVL
B2AR_PIG -----MCQPGNRSVFLAPNCSAPPDQDPQRDEAMV.VGAILVMSVLIVLAIVPNNVL
B2AR_MOUSE -----MGPHGNSDFLAPNCSAPPDHDVTPQRDEAMV.VGAILMSVLIVLAIVPNNVL
B4AR_MELGA -----MTPFPANCS.VGNCSWAAVLSRQWA.VGAA.LSITITIVIVANGLN
D1A2_EEL -----MDLNFTSVLDS.GLSBT.DSSVRLNLCGCLSSPIVSTLGNL
D1B8_CARP -----MRABRS.GAQHA.RPN.RA.ACELTRACITLWLLGNL
D5DR_FUGRU MENFNYTEPEPTPRGGVDPLRVVTAEDVPAVGV.GVSRALTCV.CAGVSTLGNL
D1B_AMPHIOXUS -----MSANTVTSPTETTANLVNNTSEASVGSCFAPNPSYAGQAVSLITIVLITLVIGNVL
5H4_CAVPO -----MDKLDANVSSKSGFSGVKRVLLTFLFLAVMAILGNL
HGPRBMY26 -----MESSFSFGVILVAVASLITATNLT

B2AR_CANFA VITATAKBERLOI.VTNVFTISLACADLVKGLAVVPFGASHILMKMVPFNEMCEFWIST
B2AR_PIG VITATAKBERLOI.VTNVFTISLACADLVKGLAVVPFGASHILMKMVPFGSFCFWIST
B2AR_MOUSE VITATAKBERLOI.VTNVFTISLACADLVKGLAVVPFGASHILMKMVPFNEMCEFWIST
B4AR_MELGA VITATAKTPRLOI.VTNVFTISLACADLVKGLVVPFGATILLSGHWVPTVVCGLWISL
D1A2_EEL VCAAVTKFRHLRSEFTVTFEVLISAVSDLLMAILVMEKVAATEVAGGWPFQSG.CINIVAE
D1B8_CARP VCATVVFRRHLRAKTHVFTISLAVSDLLMAVLVMEKVAEVAEAGGWPFQAG.CINIVAE
D5DR_FUGRU VCAAVTKFRHLRSEFTVTFEVLISAVSDLLMAVLVMEKVAEVAEAGGWPFQSG.CINIVAE
D1B_AMPHIOXUS VILAVTCHRRKMT.VTNVFTISLACADLVSGITVLPFAATLNLGWPFQSG.CDVIVSE
5H4_CAVPO VMVAVCRDRKRLKNTVTFEVLISAVSDLLVSVLMPFGAIELQVQDINMEKPOLVTRISL
HGPRBMY26 VAVAVLLIHKNDGSLCTNLNLNADTLIAVAGSLGLTGLSSPSRKTOKTCLSRMAE

B2AR_CANFA DVLGVTSASITFLCVIAVDRYVAITSPFFVQSLTKNKARNVILMVIVSGLSFLPIQ
B2AR_PIG DVLGVTSASITFLCVIAVDRYVAITSPFFVQSLTKNKARNVILMVIVSGLSFLPIK
B2AR_MOUSE DVLGVTSASITFLCVIAVDRYVAITSPFFVQSLTKNKARNVILMVIVSGLSFLPIK
B4AR_MELGA DVLGVTSASITFLCVIAVDRYVAITFAGLEALVKGRAWAVCVMAIASLISFLPIK
D1A2_EEL LKNCSTASTIKLGVSDRYVAITSPFFVQSLTKNKARNVILMVIVSGLSFLPIQ
D1B8_CARP LKNCSTASTIKLGVSDRYVAITSPFFVQSLTKNKARNVILMVIVSGLSFLPIQ
D5DR_FUGRU LKNCSTASTIKLGVSDRYVAITSPFFVQSLTKNKARNVILMVIVSGLSFLPIQ
D1B_AMPHIOXUS LKNCSTASTIKLGVSDRYVAITSPFFVQSLTKNKARNVILMVIVSGLSFLPIQ
5H4_CAVPO DVLGVTSASITFLCVIAVDRYVAITSPFFVQSLTKNKARNVILMVIVSGLSFLPIQ
HGPRBMY26 VTSAAASLTVMLITDRYLAIK.OPEVVKLVSGFVACAGLIVLVSFLGLRGE

B2AR_CANFA HMYV...ATHCGLNCAKTC.CDFEQNQVAISSVFSYFVLPLVMVVPVSVRFOVA
B2AR_PIG HMYV...ATHREALNCAVREAC.CDFEQNQVAISSVFSYFVLPLVMVVPVSVRFOVA
B2AR_MOUSE HMYV...ATHKATDCVTEETC.CDFEQNQVAISSVFSYFVLPLVMVVPVSVRFOVA
B4AR_MELGA HMYV...DGAEQAVRCYDUPRC.CDFEQNMIVVAISSVFSYFVLPLVMVVPVSVRFOVA
D1A2_EEL NHHKACTTSYFDHNGSGDLLLDNCSSINRTIYVAISSLSFSYFVLPLVMVVPVSVRFOVA
D1B8_CARP DHHKTADAGA.AEPNAS..D.ADSGSSLSRVAISSLSFSYFVLPLVMVVPVSVRFOVA
D5DR_FUGRU NHHKADNNNSAHEOG.....DGNASINRTIYVAISSLSFSYFVLPLVMVVPVSVRFOVA
D1B_AMPHIOXUS GMYV.DNQSE.BADAINVSDPLL.CIFTASTAYVAISSLSFSYFVLPLVMVVPVSVRFOVA
5H4_CAVPO GNNIGIVDLTEKRRKFNQNSNSTYGVPMVNFYVAITCSVVFYFVLPLVMVVPVSVRFOVA
HGPRBMY26 PMEQOT.....AMKGCSSFVAIPH..FVLTLSCGFFPAKLVFVFCDMLKIN

B2AR_CANFA QRLQKIDRSE.GRFHAONLSQVEQDGRSGHGHRSSK.FCLKEKALKTLGIIMGIFTL
B2AR_PIG RRLQKIDKSE.GRFHAONLSQAEQDGRSGGPHRSSK.FCLKEKALKTLGIIMGIFTL
B2AR_MOUSE KRLQKIDKSE.GRFHAONLSQVEQDGRSGHGHRSSK.FCLKEKALKTLGIIMGIFTL
B4AR_MELGA TRHVOI.GKDK.VRFLOENPLSSRGSR...WRPSSLIAKHKALKTLGIIMGIFTL
D1A2_EEL QKQTRRISALBRRA.BSAKNRHNMGNSSVETESSFRMSFKRETIRVLTLSVINGVEVC
D1B8_CARP QKQTRRISALBRRA.BHAQSR...SDRSLHRSLKTSFORRETIRVLTLSVINGVEVC
D5DR_FUGRU QKQTRRISSLBRRAQAQRAQ...SHRSTHDESALTSFKRETIRVLTLSVINGVEVC
D1B_AMPHIOXUS RDKARINALE.GRLBOENRGK.....KISLAKMKALKTLGIIMGIFTL
5H4_CAVPO KEHARQIQVLDORAGAPAE...GRPOPADOHSR.MRTETKAAKTLGIIMGIFTL
HGPRBMY26 SMHSQQRIRKSHAGAMA.....GGYRSPTT.PSDFKARTVSVLISLAE

B2AR_CANFA CWLPFFIVNIHVHVIC.....NLITKEVYILNM.VGVVNSAENFLIYCR.SPQF
B2AR_PIG CWLPFFIVNIHVHVIC.....NLITKEVYILNM.VGVVNSAENFLIYCR.SPQF
B2AR_MOUSE CWLPFFIVNIHVHVIC.....NLITKEVYILNM.VGVVNSAENFLIYCR.SPQF
B4AR_MELGA CWLPFFIVNIHVHVIC.....PLVFDOLIEUML.VGVVNSAENFLIYCR.SPQF
D1A2_EEL CWLPFFIVNIHVHVIC.....PLVFDOLIEUML.VGVVNSAENFLIYCR.SPQF
D1B8_CARP CWLPFFIVNIHVHVIC.....PLVFDOLIEUML.VGVVNSAENFLIYCR.SPQF
D5DR_FUGRU CWLPFFIVNIHVHVIC.....PLVFDOLIEUML.VGVVNSAENFLIYCR.SPQF
D1B_AMPHIOXUS CWLPFFIVNIHVHVIC.....PLVFDOLIEUML.VGVVNSAENFLIYCR.SPQF
5H4_CAVPO CWLPFFIVNIHVHVIC.....PLVFDOLIEUML.VGVVNSAENFLIYCR.SPQF
HGPRBMY26 CWLPFFIVNIHVHVIC.....PLVFDOLIEUML.VGVVNSAENFLIYCR.SPQF

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. Figure 2B

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B2AR_CANFA      R I A P O E L L C L R S S L K A Y G N G Y S N N S N G R S D V A G E H S G C H L G Q E K D S . . E L I C D P P P T
B2AR_PIG        R I A P O E L L C L R S S L K A Y G N G C S S N S N G R T D V T G E S G C Y L G E E K D S . . E R L C D A P C P
B2AR_MOUSE      R I A P O E L L C L R S S S K T Y G N G Y S S N S N G R T D V T G E P N T C Q L G Q E R E Q . . E L L C D P P C M
B4AR_MELGA      R S A P R K L L C C P P R A D R R L H A A P Q D P O H C S C A F S P R G D P M E D S K A V D P . . G H L R P C S E . V
D1A2_EEL        R K A S I L L C H H L C . . . . P G . S N A I E . I V S I N N N G A P P . . Q L V H N Q . . P K A G F S K . S C
D1BR_CARP       R R A P S S L L R C R T P V . . . . E T . V N A S N A L V S Y N R E A A S A C V N I I P N V . . V D E T L R M S Q
D5DR_FUGRU      R K V F V K M I C C H K C R G V T V G P N H A D L N Y D P V A M R L K K R G E N A N G T V N . . G D A N G K A N G
D1B_AMPHIOXUS  R R A F L I L L C C D D E R Y R R P S I L G Q T V P C S T T T I N G S T H V L R D T V E C G G O W E S C H P A A S S
5H4_CAVPO       R L Q L Y H M A L G V K K V L T S F L . . L F L S A R N G C P E . R P R E S S C H I V T I S S E F D G . . . . .
HGPRBMY26       R L Q L Y H M A L G V K K V L T S F L . . L F L S A R N G C P E . R P R E S S C H I V T I S S E F D G . . . . .

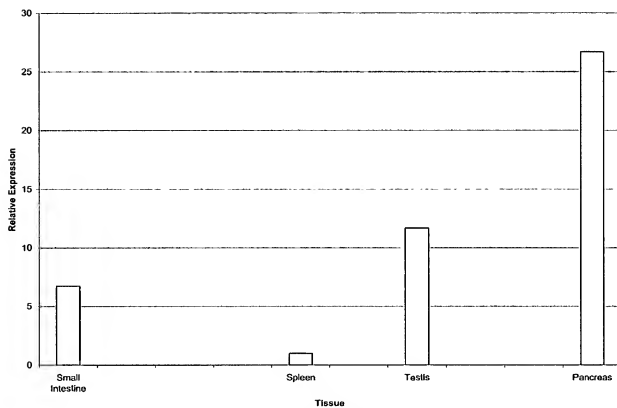
B2AR_CANFA      E . . D R C T V P S D S V D S Q G R . . . N C . . S T N D S L I . . . . .
B2AR_PIG        E G C A H R C T V P D D S T D S Q G R . . . N C . . S T N D S M I . . . . .
B2AR_MOUSE      E G F V N C C T V P S L S V D S Q G R . . . N C . . S T N D S P I . . . . .
B4AR_MELGA      Q G S G R E E N A S S H G G G H Q Q R P L G E C W L Q G M Q S M I C E Q L D E F T S T E M P A G P S V . . . . .
D1A2_EEL        . . T P K E . S N I . R H G I P H T . . I L S Q D E E L Q K K G N A I E R I S P A L S G S L D S E A D L S L D K I N P T
D1BR_CARP       . . L S R G . C D V D L D G A V H A N G I L . . . . .
D5DR_FUGRU      . . L P Q N R E D A . . . G . P N F D K V S V V S D D S R A D R N I . . L L P A I L . Q C D C E A E I S L D M V P F G
D1B_AMPHIOXUS  . . T E A G E C T S S S . . . . .
5H4_CAVPO       P L V A A Q P I D T . . . . .
HGPRBMY26       P L V A A Q P I D T . . . . .

B2AR_CANFA      . . . . .
B2AR_PIG        . . . . .
B2AR_MOUSE      . . . . .
B4AR_MELGA      . . . . .
D1A2_EEL        T Q N G Q N S T . . . . .
D1BR_CARP       . . . . .
D5DR_FUGRU      S S G P A D S F L I P G Q I Q D L G D L . . . . .
D1B_AMPHIOXUS  . . . . .
5H4_CAVPO       . . . . .
HGPRBMY26       . . . . .

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D0128 NP

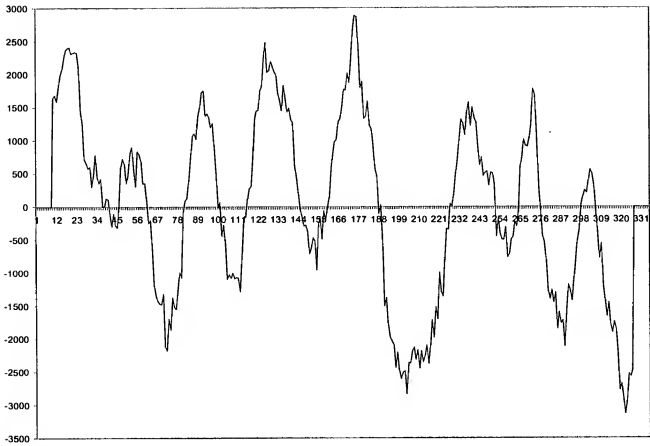
Figure 3



Note: No expression detected in brain, bone marrow, heart, kidney, liver, lung, lymph node, pituitary gland, prostate, spinal cord, or thymus)

D0128 NP

Figure 4



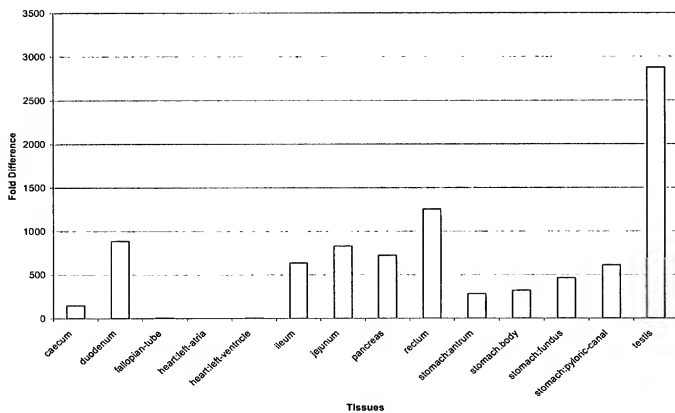
D0128 NP

Figure 5.

<u>Protein</u>	<u>SWISS PROT ID</u>	<u>Identities</u>	<u>Similarities</u>
guinea pig 5-hydroxytryptamine 4 receptor (5-HT-4) protein	gi O70528	32.18%	40.38%
Amphioxus dopamine D1/beta receptor protein	gi O96716	26.28%	35.35%
Fugu rubripes D(5)-like dopamine receptor protein	gi P53454	29.01%	37.35%
carp D1B dopamine receptor protein	gi O42317	27.22%	35.78%
eel dopamine D1A2 receptor protein	gi Q98842	29.31%	39.58%
turkey beta-4C adrenergic receptor protein	gi P43141	29.28%	38.32%
mouse beta-2 adrenergic receptor protein	gi P18762	27.52%	38.23%
pig beta-2 adrenergic receptor protein	gi Q28997	29.57%	39.94%
dog beta-2 adrenergic receptor protein	gi P54833	27.52%	38.53%

D0128 NP

Figure 6



D0128 NP

Figure 7

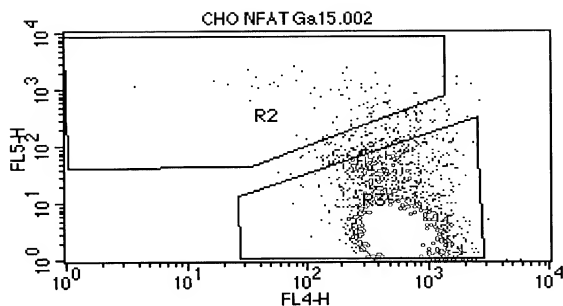
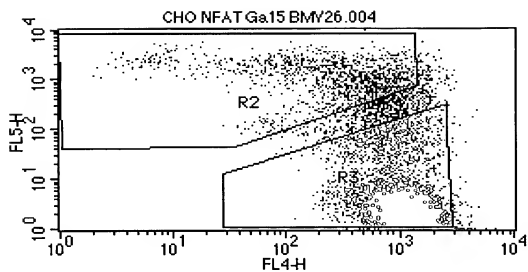


Figure 8



D0128 NP

Figure 9

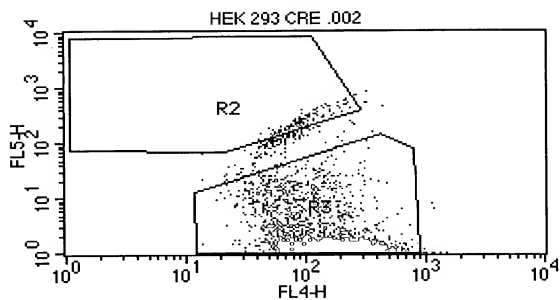
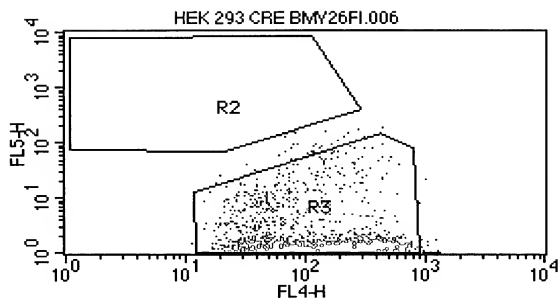


Figure 10



D0128 NP

Figure 11

Cho NFAT Ga15 Control (Fluorescent vs. Bright Field)



Cho NFAT Ga15 BMY26 (Fluorescent vs. Bright Field)

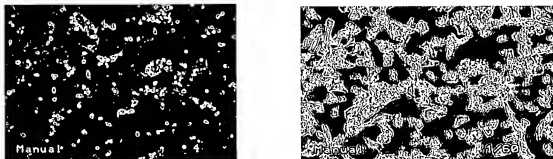
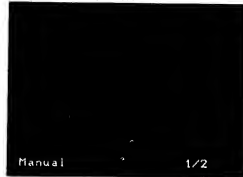


Figure 12

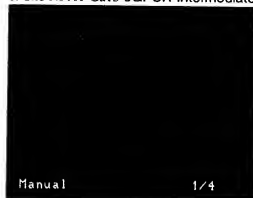
a. Cho NFAT Gal5



b. Cho NFAT Gal5 + T/P



c. Cho NFAT Gal5 oGPCR-Intermediate



d. Cho NFAT Gal5 oGPCR High

